Exhibit 1

***Kogut, Restivo and Halstead v. Nassau County, et al.*, No. CV06-6695 (JS) (WDW)**

REPORT

My background

My name is Max M. Houck. I hold a Bachelor's of Science in Anthropology and a Master's in Anthropology from Michigan State University; those degrees were awarded in 1984 and 1988, respectively. My Master's coursework emphasized forensic anthropology, human identification, decomposition, and examination of trauma to the skeleton. I have a Ph.D. in Applied Chemistry from Curtin University in Perth, Western Australia. My research topic was developing a set of foundational principles for forensic science and using supply chain mechanics to establish evidentiary significance. My Ph.D. was awarded *summa cum laude* (Chancellor's Commendation) in 2010. My career has been spent in forensic science, forensic anthropology, and trace evidence analysis.

I have worked in the private sector at Oxford Instruments from 1989 to 1992 as an Applications Specialist. I then work at a medical examiner's office in Fort Worth, TX as a trace evidence analyst and a forensic anthropologist. I instituted the forensic anthropology section in the laboratory, including recovery of remains. While at that office, I acted as the coordinating anthropologist for the Branch Davidian Investigation, in Waco, TX. From 1992 to 2001, I worked at the Federal Bureau of Investigation Laboratory Division as a Physical Scientist assigned to the Trace Evidence Unit. While there, I worked over 800 cases in trace evidence and anthropology and testified in local, state, and federal courts for the prosecution and the defense. I was the Chairman of the Scientific Working Group for Materials Analysis, which included the subgroup on Hairs led for a time by Nicholas Petraco. I was the Technical Liaison Manager with outside federal agencies and a Research Associate at the Department of Anthropology at the Smithsonian Institution. In September and October of 2001, I assisted DoD with identification of victims of the 9/11/01 attack on the Pentagon. In January of 2002, I became Director of the Forensic Science Initiative (Research) at West Virginia University, a position I held until my departure in 2011. I lead and directed my staff in the administration of $32 million in research and resource initiatives for forensic science. We developed and conducted educational and professional resources in forensic science and forensic business and economic applications and research. I was a lecturer in Forensic and Investigative Sciences Program at WVU as well as being appointed Director of Forensic Business Research and Development in the College of Business and Economics. Currently, I am Principal Analyst in the Forensic Enterprise Division at ANSER, a division of Analytic Services, Inc., a non-profit policy and

strategy institute in Arlington, VA. I am also an adjunct Associate Professor at American University's Washington College of Law in Washington, DC.

I am a Fellow of the American Academy of Forensic Science, a Senior Member of the American Association of Textile Chemists and Colorists, a member of the American Statistical Association, a member of the American Association for the Advancement of Science, an Academic Member of the American Society of Crime Laboratory Directors, and a member of the American Chemical Society. I have served on numerous professional and advisory committees, including the American Society of Crime Laboratory Directors, Training and Education Committee (2011-present), the Executive Office of the President of the United States, National Science and Technology Council, Committee on Science, Subcommittee on Forensic Science, Education, Ethics, and Terminology Interagency Working Group (2010-2011), the NIST Human Factors in Latent Print Analysis as Co-Chair (2009-2010), the Interpol Forensic Science Symposium Planning Committee (2008-present), the Technical Working Group on Education and Training in Forensic Science, and the ASTM Committee E 30 on Forensic Sciences.

I have many publications in the forensic sciences, including 10 books, numerous book chapters, and dozens of peer-reviewed journal articles, several of which involve the forensic examination of hairs I have given hundreds of presentations at professional conferences and to committees on forensic science, education, business practices of forensic laboratories, philosophy of forensic science, and my research areas of hairs, fibers, and forensic anthropology. I have also taught dozens of short courses in forensic science, trace evidence, leadership, expert testimony, grants, and visualization of data.

A list of my publications in the last 10 years follows:

Houck, M.M., Crispino, F., and McAdam, T. *The Science of Crime Scenes*. Elsevier: Amsterdam. *In Press*.

Houck, M.M. and Bowen, R.T. "Teaching Ethics," in *Ethics in Forensic Science*, Downs, J. and Swienton, A. (eds). Elsevier: Amsterdam. 2012.

Houck, M.M. and Siegel, J.A. *Fundamentals of Forensic Science*, 2nd edition. Academic Press, San Diego. 2010.

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Houck, MM, *Science vs Crime*, Facts on File, New York. 2009.

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Houck, MM, "My Roomate is Using the Refrigerator", in Houck, MM (ed) *Trace Evidence Analysis: More Cases from Mute Witnesses*, Academic Press: San Diego, 2003.

Kobus, H., Houck, M.M., Speaker, P., Riley, R. and Witt, T. 2011. "Managing Performance in the Forensic Sciences: Expectations in Light of Limited Budgets," *Forensic Science Policy and Management* (2): 1-8.

Mnookin, J., Cole, S., Fisher, B., Houck, M., Inman, K., Kaye, D., Koehler, J., Langenburg, G., Risinger, M., Rudin, N., Siegel, J. and Stoney, D. 2010. "The need for a research culture in the forensic sciences," *University of California Law Review* (58) 725-779.

Houck, M.M. and Boyle, J. 2010. "A content analysis of fingerprint literature for educational curricula," *Science and Justice*, 50: 123-126.

Houck, MM, Riley, R, Speaker, P and Witt, T. 2009. "FORESIGHT: A business approach to improving forensic services" *Forensic Science Policy and Management* 1(2): 85-95.

Vatsa, M., Singh, R., Noore, A. and Houck, M. 2009. "Quality-augmented fusion of level-2 and level-3 fingerprint information using DSm theory" *International Journal of Approximate Reasoning* 50: 51-61.

Houck, MM. 2009. "Is Forensic Science a Gateway for Women into Science?" *Forensic Science Policy and Management* 1(1): 65-69.

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Houck, MM and Quarino, L. 2008. "Saving Us From Ourselves: Recreating Forensic Science," *Forensic*

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Noore, A, Vatsa, M, Singh, R, Morris, K., and Houck, M. 2007 "Enhancing security of fingerprint images," *Journal of Documents and Identity* 22: 3-5.

Noore, A, Singh, R, Vatsa, M. and Houck, MM. 2007. "Enhancing security of fingerprints through contextual biometric watermarking," *Forensic Science International*, 169: 188-194.

Houck, MM, Kranacher, M, Morris, B, Riley RA, Robertson, J, and Wells, JT. 2006. "Forensic Accounting as an Investigative Tool", *The CPA Journal*, 76 (8): 68-69.

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Asbaugh, D and Houck, MM (2005) "Fingerprints and Admissibility: Friction, Ridges, and Science", *The Canadian Journal Of Police & Security Services Practice, Policy & Management*, 3(2), pages not available.

Houck, MM. (2005) DNA and the criminal justice system (book review), *The Journal of Clinical Investigations* 115 (6): 1398.

Houck, MM, and Bisbing, RE, (2005) Forensic Human Hair Examination and Comparison in the 21st Century, *Forensic Science Review*, V17, N1: 51-66.

Houck, MM and Bowen, R (2005) An Argument for Microscopy: A Review of Forensic Microscopy, *Forensic Science Review*, V17, N1: 1-16.

Houck, MM (2005) Forensic Fiber Examinations and Analysis, *Forensic Science Review*, V17, N1:29-50.

Houck, MM (2005) Editor's Introduction, *Forensic Science Review* V17, N1.

Noore, A, Tungala, N, and Houck, MM. (2004) Embedding biometric identifiers in 2D barcodes for improved security, *Computers and Security*, 23: 679-686.

Houck, MM, (2004) The Nature of Physical Evidence, *Law Enforcement Technology*, October, 124-133.

Houck, MM, Bisbing, RE, Watkins, T, and Harmon, RE, The Science of Forensic Hair Comparisons and the Admissibility of Hair Comparison Evidence: *Frye* and *Daubert* Reconsidered, on-line at: www.modernmicroscopy.com, 02 March 2004.

Houck, MM, 2003, Inter-comparison of unrelated textiles, *Forensic Science International*, 135(2): 146-149.

Ubelaker, DH, and Houck, MM, 2002, Utilization of Radiocarbon Dating and Paleontological Extraction Techniques in the Analysis of a Human Skull in an Unusual Context, *Forensic Science Communications*, V4, N4, available: www.fbi.gov.

Houck, MM, and Budowle, B, 2002, Correlation of Microscopic and Mitochondrial DNA Analysis of Hairs, *Journal of Forensic Sciences*, V45, N5: 1-4.

I have not been required or requested to testify in a trial or deposition in the last 4 years.

Decomposition of hairs

Decomposition is the breaking down of organic material into components or forms of matter. The process of decomposition can be described as happening through two processes of chemical activity.[1] The first is autolysis, which is the degradation and eventual breaking apart of cells by their own enzymes. The second is putrefaction, which is the decomposition of proteins and results in the loss of structural integrity of and cohesion between tissues; this process leads to the dissolution of larger anatomical structures, such as tissues, muscles, and organs in animals. Once an animal's heart stops beating, these chemical alterations begin and cause changes in the body's acidity (pH); microorganisms, such as bacteria, on and in the body, begin to digest the tissues, causing a loss in cellular integrity. As the cells break down, more enzymes are released and the process of decomposition progresses and spreads to surrounding cells and tissues, eventually encompassing all of the soft tissues of the body.[2]

Hair follicles and hair roots are not immune to the process of decomposition and the changes seen are based in part on the growth cycle of hairs. Hairs[3] have three growth phases: active (or anagen), transitional (catagen), and resting (telogen); hair growth is cyclical and each hair goes through all three phases in the order described. As hairs actively grow (anagen phase) in the follicle (the structure in the skin—or epidermis—containing the growing hair), they are generated at the base of the follicle and then are pushed upwards towards the surface of the skin by the addition of new cellular material at the base. The hair is soft at the bottom of this base and gradually hardens as it progresses outward; the hardening

[1] Forbes, S.L. 2008. "Decomposition Chemistry in a Burial Environment". In M. Tibbett, D.O. Carter. *Soil Analysis in Forensic Taphonomy*. CRC Press: Boca Raton, FL. pp. 203–223.
[2] Pinheiro, J. 2006. "Decay Process of a Cadaver". In A. Schmidt, E. Cumha, J. Pinheiro. *Forensic Anthropology and Medicine*. Humana Press: New York. pp. 85–116.
[3] This section taken generally from Montanga, W. and Ellis, R. 1958. *The Biology of Hair Growth*. Academic Press: San Diego and Swift, J.A. 1997. "Morphology and Histochemistry of Human Hair" In *Formation and Structure of Human Hair*. P. Jolles, H. Zahn, and H. Hocker, eds. Pp. 149-176. Boston: Birkhauser Verlag.

process is called keratinization, after the material which hair is made of (keratin). Thus, one can speak of "soft" keratin at the base of an anagen hair eventually becoming "hard" keratin at the level of the skin. Hairs in anagen phase can actively grow for several years, depending on health and genetics. Follicles transition to the catagen phase (for only a few weeks) before shutting down growth for a time. The hair then moves into the resting telogen phase, where the entirety of the hair, including the base or root, keratinizes and becomes hard. The hair is then shed, signaling the follicle to reactivate and begin the anagen phase again.

In decomposition, hairs that were actively growing (anagen or early catagen) until the time of death go through changes in their root ends related to the decomposition of the surrounding skin and follicle. One of the phenomena observed in these former anagen or early catagen hairs is called "putrid root" or "postmortem root banding" [4,5,6,7]. The phenomenon is seen as "an opaque ellipsoidal band which appears to be composed of a collection of parallel elongated air spaces" near the root of a hair, appearing as a dark or blackened band in the hair shaft[8]. Although the exact mechanisms of origin for postmortem root banding are not known, it has been suggested that the banding is a result of the "soft" keratin at the base of an anagen or early catagen hair being affected by the putrefaction of the surrounding tissue.[9] Hairs from an individual tend to decompose at the same rate and the statistical relationship between postmortem root morphology and postmortem interval (time after death) is significant.[10]

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[4] Linch, C. A. and Prahlow, J.A. 2001. "Postmortem Microscopic Changes Observed at the Human Head Hair Proximal End", *Journal of Forensic Sciences* 46(1):15-20.

[5] Tafaro, J. T. 2000. "The Use of Microscopic Postmortem Changes in Anagen Hair Roots to Associate Questioned Hairs with Known Hairs and Reconstruct Events in Two Murder Cases", *Journal of Forensic Sciences* 45(2):495-499.

[6] Petraco, N., Fraas, C., Callery, F.X., and DeForest, P.R. 1988. "The Morphology and Evidential Significance of Human Hair Roots", *Journal of Forensic Sciences* 33(1):68-76.

[7] Seta, S., Sato, H.. Yoshino, M., and Miyasaka, S.. 1984. "Morphological Changes of Hair Root with Time Lapsed After Death" in *Proceedings,* 10th Triennial Meeting of the International Association of Forensic Sciences, Section on the Characterization of Human Hair, Oxford, UK, 18-25 September.

[8] Petraco et al, 1988, page 73.

[9] Linch and Prahlow, 2001.

[10] P = 0.011; see Collier, J.H. 2005. *Estimating the Postmortem Interval in Forensic Cases through the Analysis of Postmortem Deterioration of Human Head Hair*. Master's Thesis, Louisiana State University.

It has been documented that days must pass before postmortem root banding presents itself in hairs, with the minimum being 7 or so days;[11] in one clinical study, no postmortem root banding was seen in the six "youngest" cases, ranging from half a day to four days.[12]

The transformation of the putrid root *only* occurs in roots that remain in the scalp of a decomposing body; the changes do not occur if the hair is plucked (or shed) prior to death and allowed to deteriorate[13]. In a recent study conducted by the FBI Laboratory Division[14], 600 hairs(97.5% of which were in the anagen phase when collected from living individuals) were studied for postmortem changes. Two-hundred and fifty hairs were stored in vehicles or indoors on a windowsill from 9 to 230 days, 250 hairs were stored outdoors on the ground surface in shaded and non-shaded areas from 7 to 106 days, and 100 hairs were submerged in water or buried in potting soil from 15 to 100 days. In this study, *no hairs stored indoors on the windowsill and no hairs stored in the vehicles exhibited characteristics of decomposition* and *no hairs in this study exhibited characteristics of postmortem banding.* Thus, for a hair to present postmortem root banding based on the published literature, it must have been:

- in the anagen or early catagen phase of growth prior to death,
- in the skin during the time of decomposition, and
- in the decomposing skin for a minimum of 7 days.

Thus, the decomposition of hair roots and postmortem root banding are part of the process of establishing postmortem interval (PMI).

Offered timeline

Based on the timeline offered by the prosecution during the criminal proceedings, It would not be possible for any of Ms. Fusco's hair left in the van during the commission of the crimes to have

[11] Linch and Prahlow, 2001; Collier, 2005.
[12] Linch and Prahlow, 2001.
[13] Tafaro, ibid.
[14] Shaw, S., Otterstatter,L., Shegogue, C., Lowe, K., Koch, S., and Friedman, J. 2012. "The Microscopic Characteristics of Antemortem and Postmortem Hairs at the Root End," presented at the American Academy of Forensic Sciences annual conference, Atlanta, GA. Abstract A158.

developed postmortem root banding. I understand that the prosecution offered the following timeline of events in their theory of the crime:

- At approximately 10 pm on the night of November 10, 1984, while driving in Mr. Restivo's blue van, plaintiffs Restivo, Halstead and Kogut encountered Ms. Fusco as she walked on a section of Merrick Road in Lynbrook that runs alongside a cemetery;
- Fusco got in the van and was attacked by the plaintiffs, one of whom raped her as the van was driven a very short distance into the cemetery bordering the street where she was picked up, and another who raped her after the van arrived in the cemetery;
- After the rapes, Fusco was carried out of the van while alive but unconscious and placed on a blanket on the ground of the cemetery, where she was strangled to death;
- Immediately thereafter, her body was wrapped in the blanket and placed inside the van;
- The plaintiffs then drove a few blocks, stopped the van, removed Ms. Fusco's body, and carried her body a short distance to a wooded area known as "the Fort", where they dumped it.
- All of these events are alleged to have occurred in perhaps less than an hour, and the prosecution contended at the criminal trial that the questioned hair samples (2 Q8 hairs and 1 Q4 hair) purportedly collected from the blue van and later identified as having come from Ms. Fusco were inadvertently left behind during the course of the above-noted events.

Based on the known and documented scientific clinical studies on postmortem root banding relating to its timing, description, appearance, and conditions for existence, there is no known mechanism or reasonable explanation for postmortem root banding to appear in Ms. Fusco's hairs that were allegedly left in the blue van in the above scenario.

I hereby declare under penalty of perjury that the foregoing is true.

Sworn this 16th day of March, 2012
Arlington, VA

Max M. Houck

Max M. Houck *202.670.4661 max.houck@gmail.com*

Principal Analyst, Forensic Enterprise Division, *2011 (April)--present, Analytic Services, Inc. (ANSER), Arlington, VA*

- Thought-leader in forensics and biometrics; National-scale strategy for forensic science
- Assistance to various national projects and programs in forensic-related areas
- **Top Secret Clearance, 2011-2016**

Vice President and Director of Forensic Services, *2011--present, Forensic and Intelligence Services, LLC, Arlington, VA*

- Contractor providing subject matter expertise in intelligence analysis, forensic science, business and process improvement, and professional communications
- Woman/Minority-owned small business

Adjunct Associate Professor, *2011--present, American University Washington College of Law, Washington, DC.*

- Instructor for course in scientific evidence; additional academic duties

Director, Forensic Science Initiative (Research); Director, Forensic Business Research and Development (Business and Economics), *2001–2011 (April), West Virginia University, Morgantown, WV*

- Direction of $32 million in research and resource initiatives for forensic science
- Developing educational and professional resources in forensic science
- Developing forensic business and economic applications and research
- Lecturer in Forensic and Investigative Sciences Program, WVU
- H-Index: 13 (highest H-Index forensic author = 43)

Physical Scientist, Trace Evidence Unit, Laboratory Division, *1994–2001, Federal Bureau of Investigation, Washington, D.C.*

- Worked over 800 cases in trace evidence and anthropology; Testified in local, state, and federal courts for prosecution and defense
- Chairman of the Scientific Working Group for Materials Analysis
- Technical Liaison Manager with outside federal agencies
- Research Associate at Department of Anthropology, Smithsonian Institution
- **Assisted DoD with identification of 9/11/01 Pentagon victims**

Criminalist II, *1992-1994, Tarrant County Medical Examiner, Fort Worth, TX*

- Trace evidence analysis; Instituted forensic anthropology department in laboratory, including recovery of remains; **Coordinating anthropologist for the Branch Davidian Investigation, Waco, TX**

Applications Specialist, *1988-1992, Oxford Instruments, Inc., Madison, WI*
Electron Microscopy Technician, *1987-1988, Michigan State University, East Lansing, MI*
Archaeologist and Research Assistant, *1982-1986, Michigan State University, East Lansing, MI*

Education

- 2010 Ph.D., Applied Chemistry, Curtin University, Perth, Australia: *Foundational Principles of Forensic Science: Using Supply Chains as a Basis for Evidentiary Significance*. Chancellor's Letter of Commendation.
- 1988 M.A., Anthropology, Michigan State University, *Individualization of Toolmarks in Bone*
- 1984 B.S., Anthropology, Michigan State University, Outstanding Senior in Anthropology

Professional memberships

- Fellow, American Academy of Forensic Science
- Senior Member, American Association of Textile Chemists and Colorists
- American Statistical Association
- American Association for the Advancement of Science
- American Society of Crime Laboratory Directors, Academic Member
- American Chemical Society

Awards

- Best Oral Presentation, Australian-New Zealand Forensic Science Society Symposium 2010, Management and Quality Section, "The FORESIGHT Project: Collaborative Benchmarking for Effectiveness, Efficiency, and Quality"
- Mary E. Cowan Outstanding Service Award, American Academy of Forensic Sciences, 2009
- 100 Most Influential (with Clifton Bishop), *Dominion Post*, Morgantown, WV, 2004
- American Society for Testing and Materials, Forensic Sciences Award, 2001
- Quality Award, FBI Laboratory, 1999

Editorial work

- Founding Co-Editor (with Jay Siegel), *Forensic Science Policy and Management*
- Editorial Board, *Science and Justice*
- Editorial Board, *Journal of Forensic Sciences*

Committees

- American Society of Crime Laboratory Directors, Training and Education Committee (2011-present).
- Advisory Member, Executive Office of the President of the United States, National Science and Technology Council, Committee on Science, Subcommittee on Forensic Science, Education, Ethics, and Terminology Interagency Working Group (2010-2011)
- NIST Human Factors in Latent Print Analysis, Co-Chair (2009-2010)
- Interpol Forensic Science Symposium Planning Committee (2008-present)
- Forensic Educational Program Accreditation Commission (FEPAC), Chairman (2005-2011)
- Forensic Educational Program Accreditation Commission (FEPAC) 2004
- European Academy of Forensic Sciences Symposium Scientific Committee (2005-2006)
- National Academies Committee on Educational Paradigms for Homeland Security (2004)
- Business Conference Advisory Panel, Nemacolin Woodlands Resort and Spa (2003-2004)
- National Library of Medicine Planning Panel, History of Forensic Science exhibit, 2002-2003
- Technical Working Group on Education and Training in Forensic Science
- ASTM Committee E 30 on Forensic Sciences
- Imaging Technology Coordination Committee (FBI), Chairman, Standards and Practices
- AEGIS Council, Support (Non Agent) Employee Advisory Committee to the FBI Director (1997 1999). Addressed and mitigated FBI employee concerns with administration and reported to Director Freeh.
- AEGIS Representative for Division 7 (Laboratory), 1995 1997, 1997 1999. Represented Division 7 employee concerns to AEGIS Council.
- Technical Advisory Panel, Revision of Standard Reference Collection of Forensic Science Materials: Status and Needs (1977), through Office of Law Enforcement Standards (OLES) and National Institute of Technology and Standards (NIST).

Keynote addresses and Invited Lectures

- Florida and Georgia Chapters of the International Association for Identification, Panama Beach City, FL, October, 2010.
- Forensic science as a gateway for women into science, Oxford Round Table invited presentation, July, 2009.

- Forensic science, CSI, and education, West Virginia Science Teachers Association, October, 2008.
- Presentations to the National Academies of Science "Needs of Forensic Science" Committee on forensic science education and forensic hair comparisons
- Crime Scene Investigation, NASA Goddard Engineering Colloquium, Goddard Space Flight Center, Greenbelt, MD, November 2005
- The Role of Forensic Science and Academia, Educational Session, International Association of Forensic Sciences, 2005, Hong Kong, China.
- Forensic Science and Business, 2005, West Virginia University Alumni Association, Cleveland, OH
- Forensic Science is History, 2004 Combined Meeting of the Southern, Midwestern, MidAtlantic Associations of Forensic Scientists and the Canadian Society of Forensic Scientists, Orlando, FL, September.
- Science and the Law: Nexus of Conflict and Resolution, 2003 Forum on Crime Laboratory Management for Criminal Justice Agencies, The Performance Institute, Arlington, VA, July 30-31.
- Making Legal Evidence Count, 2003, Max Planck Institute Summer Course in Bounded Rationality, Berlin, Germany.

Books and book chapters

Houck, M.M., Crispino, F., and McAdam, T. *The Science of Crime Scenes*. Elsevier: Amsterdam. *In Press*.

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Ryland, S and Houck MM, "Only Circumstantial Evidence," in Houck, MM (ed), *Mute Witnesses: Trace Evidence Analysis*, Academic Press: San Diego, 2001.

Wiggins, K and Houck, MM, Introduction, in Houck, MM (ed), *Mute Witnesses: Trace Evidence Analysis*, Academic Press: San Diego, 2001.

Siegel, J and Houck, MM, Chapter 36: Forensic Textile Fiber Analysis, In *Forensic Sciences* (3 vol.), C. Wecht, ed., 2001.

Houck, MM, 1998, Skeletal Trauma and the Individualization of Knife Marks in Bone, in *Forensic Osteology*, 2nd ed., Reichs K (ed.), 410 424.

Journal Publications

Crispino, F., Ribaux, O., Houck, M., and Margot, P. 2011. "Forensic science--A true science?" *Australian Journal of Forensic Sciences*, in press.

Houck, M., Fleming, S., Speaker, P., Riley, R., 2011. "The balanced scorecard: Sustainable performance assessment for forensic laboratories," *Science and Justice*, in press.

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Thesis and Dissertation Committees

Chair

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Reader

- Brooks, E.,2007. *An appraisal of the use of numerical features in the forensic examination of hair.* MS, School of Health Sciences. University of Canberra, Australia.
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2003-RC-CX-K001 $8 million
2008-DN-BX-K223 $3.57 million
2009-DI-BX-K012 $4 million
2010-DI-BX-K011 $4 million

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ESTIMATING THE POSTMORTEM INTERVAL IN FORENSIC CASES THROUGH THE ANALYSIS OF POSTMORTEM DETERIORATION OF HUMAN HEAD HAIR

A Thesis

Submitted to the Graduate Faculty of the
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In

The Department of Geography and Anthropology

By
Jamie Hughes Collier
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ABSTRACT

Establishing the postmortem interval (PMI) of a decedent is one of the most important responsibilities a forensic investigator may face. An accurate PMI may aid in the identification of not only the victim, but also a suspect. Although many methods for determining time since death have been proposed, there is still a need to establish more reliable dating techniques. This study determines whether head hair from an individual deteriorates uniformly and if so, what association cuticle damage, fungal growth, and changes in proximal end morphology may have with PMI.

Fifteen to 25 scalp hairs were pulled from nine cadavers located in the outdoor field of the University of Tennessee Anthropological Research Facility. In addition, 15 hairs were pulled from a living 59-year-old, Caucasian male to be used as a control. Each case was placed in a category for cuticle damage, fungal growth, and proximal end morphology through the use of microscopic observations. Chi-square tests were used to determine whether head hair from the same individual deteriorates uniformly, what association cuticle damage, fungal growth, and changes in proximal end morphology may have with PMI, and what association cuticle damage, fungal growth, and changes in proximal end morphology have with each other.

This study demonstrates that head hair from the same individual deteriorates uniformly. In addition, fungal growth and changes in proximal end morphology have a significant association with PMI; conversely, cuticle damage and PMI have a nonsignificant relationship. A significant association exists between fungal growth and changes in proximal end morphology. On the other hand, the relationships between cuticle damage and fungal growth, and cuticle damage and changes in proximal end morphology were not significant.

Utilized in conjunction with other dating methods, the observations of fungal growth and changes in proximal end morphology of human head hair may prove beneficial in estimating a PMI.

CHAPTER 1: INTRODUCTION

One of the most important goals a forensic investigator faces is establishing the postmortem interval (PMI) of a deceased individual. Correctly establishing time since death can mean the difference between serving justice and allowing a criminal to remain free. While the forensic pathologist's and forensic entomologist's duty is typically to determine time since death in cases with shorter postmortem intervals, the forensic anthropologist determines time since death in cases with longer postmortem intervals. Often a forensic anthropologist can establish PMI by identifying the stage of decomposition of the decedent (Bass 1997; Clark et al. 1997; Haglund and Sorg 1997; Buchan and Anderson 2001). At times, the numerous variables involved in this method make ascertainment of an accurate PMI difficult. Even experienced anthropologists can incorrectly estimate time since death (Bass 1997; Ubelaker 1997). Decomposition rates are not the only method for determining PMI. Over a dozen different methods for determining longer postmortem intervals have been documented, but there is still a great need to establish more reliable dating methods (Buchan and Anderson 2001).

Although many researchers have examined the deteriorative effects of human head hair on making forensic comparisons (Lasko 1984; Serowik and Rowe 1986; Kundrat and Rowe 1988; Petraco et al. 1988; DeGaetano et al. 1992; Kupferschmid et al. 1994; Tafaro 2000; Linch and Prahlow 2001), few have investigated the idea that the biodeterioration of human head hair could be used in determining PMI (Lasko 1984; Linch and Prahlow 2001). Observations of the different characteristics of the deterioration of human head hair may be beneficial in determining PMI.

Cuticle Damage

The cuticle is one of three layers of human hair; it is the first line of defense in protecting against environmental deterioration. The cortex, the layer below the cuticle, makes up the bulk of the hair and is responsible for the strength of the hair. The innermost layer, the medulla, contains large intercellular spaces and can be absent, fragmented, or continuous (Montagna 1962; Robbins 1988; Swift 1997). The cuticle is composed of overlapping cells, or scales, which begin at the proximal end of the hair and point distally, similar to the shingles on a roof (Robbins 1988). As with a roof protecting the interior of a building, the cuticle is responsible for protecting the cortex and medulla from the environment. In addition, the cuticle scale pattern can aid in identifying animal species (Robbins 1988; Kupferschmid et al. 1994). Kupferschmid et al. (1994) examined the effect hair deterioration had on making successful species identification. Kupferschmid et al. (1994) observed progressive deterioration of cuticle scales of hairs buried in soil and immersed in water over an eight-week period. In addition to the deterioration of the cuticle, Kupferschmid et al. (1994) also witnessed increased fungal attack on the shaft of the buried hairs.

Fungal Growth

The destruction of hair by keratinophilic and non-keratinophilic fungi has been well documented (Griffin 1960; English 1963, 1965, 1969, 1976). Griffin (1960) identified over 30 genera of fungi on hair in contact with soil from three different locations. All species of fungi are not present on the hair at the same time. Griffin (1960:594) described an order of succession where the hair "will first be occupied by fungi with high competitive saprophytic ability able rapidly to utilize the less complex nutrients of the substrate." The hair is then progressively occupied by fungi with less competitive saprophytic ability. Keratinophilic fungi

are the final fungi to appear utilizing keratin, the most resistant part of the substrate (Griffin 1960). Although a keratinase enzyme has never been isolated from keratinophilic fungi, researchers speculate that an enzyme is the primary mode for the keratinophilic fungi to penetrate the hair cuticle. Even though the non-keratinophilic fungi lack the keratinase enzyme, some are able to penetrate the shaft by mechanical pressure of a boring hypha, whose sole purpose is mechanical and not nutritive (English 1965).

Lasko (1984) was one of the first to investigate the importance of the deterioration of human hair with respect to forensic science. Although Lasko (1984) explained many characteristics of the deterioration of head hair, the presence of fungal growth on the hair was not mentioned. On more than one occasion Lasko (1984) described the increasing amount of debris on the shaft of the hair as the period of study progressed. The aforementioned debris could be evidence of a fungal occupation. Serowik and Rowe (1986) and Kundrat and Rowe (1988) described the deterioration of hair buried in potting soil and agricultural soil, respectively. Fungal tunnels, which are produced by thin hyphae tunneling perpendicular through the shaft of hair, were witnessed in both experiments only one month after burial. Similarly, fungal tunnels were observed in hairs from a case where the victim had been buried for three weeks (DeGaetano et al. 1992). Unlike Serowik and Rowe (1986) where the hair was cut from the scalps of living subjects and Kundrat and Rowe (1988) where the hair was plucked from the scalps of living subjects, the hairs described in DeGaetano et al. (1992) were in direct association with a decomposing body. Although DeGaetano et al. (1992) described the deterioration to the shaft of the head hair, no mention was made of the changes witnessed in the proximal end morphology.

Changes in Proximal End Morphology

Changes in proximal end morphology occur during the anagen (active growth) and catagen (transitional) stages of hair growth to the decomposing or putrid root (Petraco et al. 1988; Tafaro 2000; Linch and Prahlow 2001). The transformation of the putrid root only occurs in roots that remain in the scalp of a decomposing body; the changes do not occur if the hair is plucked prior to death and allowed to deteriorate (Tafaro 2000). The occurrence of postmortem root banding was first described by Petraco et al. (1988:73) as "an opaque ellipsoidal band which appears to be composed of a collection of parallel elongated air spaces" along the proximal portion of a hair shaft. Although the mechanisms involved in creating a postmortem root band are unknown, Linch and Prahlow (2001) suggested that the change occurs around the keratogenous zone of the hair root. Petraco et al. (1988) suggested that the brush-like appearance of some putrid roots is due to a fracture at the root band, leaving the proximal end of the hair in the scalp. Finding brush-like proximal ends may be indicative of a root with prolonged exposure and may one day help determine a new method for discovering PMI (Petraco et al. 1988). On the other hand, Linch and Prahlow (2001) believed that brush-like proximal ends are not due to prolonged exposure, but from being present in a moist scalp area. Hard keratin point proximal ends, a third type of putrid root morphology, most likely arise in dry scalp areas according to Linch and Prahlow (2001).

Purpose of Research

While Linch and Prahlow (2001) pointed out that they do not believe that proximal end morphology can be an indicator of PMI, they did not explain why their three oldest cases, ranging from 30 days to 16 years, only exhibited hard keratin point and brush-like proximal end morphology. In addition, their six youngest cases, ranging from half a day to four days, only

exhibited normal antemortem roots. Furthermore, their cases had a huge gap in PMI. The oldest two cases had PMIs of 90 days and 16 years.

While several studies examined the deterioration rate of buried and submersed head hair (Lasko 1984; Serowik and Rowe 1986; Kundrat and Rowe 1988; DeGaetano et al. 1992; Kupferschmid et al. 1994), only one study observed the deteriorative rate of head hair found on the ground surface (Lasko 1984). Unfortunately, Lasko (1984) spent little time discussing the deterioration found in this instance. None of the long-term studies of the deterioration of head hair (Lasko 1984; Serowik and Rowe 1986; Kundrat and Rowe 1988; Kupferschmid et al. 1994) took into consideration the effects decomposing tissue could have on the deterioration of hair. According to Janaway (2002), the decomposition of wool samples, which are composed primarily of the same constituents as human hair, was greatly retarded when in association with decomposing tissue. Janaway (2002:398) believed that the decomposition was impeded because "the actively decomposing soft tissue form[ed] a semi-liquid, anaerobic environment within which only a very specialized microflora can operate."

None of the previous studies examined the characteristics (i.e., cuticle damage, fungal growth, and changes in proximal end morphology) that can occur during the deterioration of human head hair. This study investigates whether head hair from an individual deteriorates uniformly and if so, what correlation cuticle damage, fungal growth, and changes in proximal end morphology may have with PMI.

CHAPTER 2: MATERIALS AND METHODS

The study began late summer of 2003 in Knoxville, Tennessee at the University of Tennessee Anthropological Research Facility (ARF), which is under the direction of Dr. Richard Jantz. Fifteen to 25 scalp hairs were pulled from each of nine cadavers located in the outdoor field. The nine cadavers were selected for sampling because they still contained scalp hair. Due to the fact that ARF has contained many cadavers in the past, hair was not introduced into the sample unless it was attached to scalp tissue in proximity of its cadaver. In some cases, individual strands of hair were not easy to obtain due to the presence of adipocere on the scalp and hair. In these instances, several hairs were pulled at once. On the opposite end of the spectrum, less than 15 hairs could be found on a balding individual. In this case, all of the head hair that was available was pulled. The hairs were pulled from several areas on the scalp to ensure a random sampling of hair. The samples were placed in labeled Ziploc bags. A digital picture was taken of each cadaver for quick reference of environmental conditions and state of the remains. In addition, sample number, age, sex, race, cause of death, date of death, and date of deposition were recorded for each case (Appendix).

Upon arrival in Baton Rouge, Louisiana, the sample hairs were stored in the Louisiana State University (LSU) Forensic Anthropology Laboratory. A control case of head hair was obtained before microscopic examination began. Fifteen scalp hairs were pulled from a 59-year-old, Caucasian male to be used for the control. The control hairs were stored in the same type of Ziploc bag as the previous collected hairs. The sample of control hair was brought to the LSU Forensic Laboratory for preparation for microscopic examination.

All hair was rinsed in room temperature tap water to remove the adipocere on several of the samples. Even though some hair was free of adipocere, such as the control, all hair was rinsed to ensure uniformity. To prevent loss of the hair during rinsing, a fine sieve was

constructed using a pantyhose covered wire strainer. The sieve allowed water to pass through, but not hair. Each sample of hair, including the control, was placed on the sieve and gently rinsed under low water pressure. In several cases, forceps were used to tease the strands of hair that were "adhered" together by the adipocere. The greatest effort was taken to be gentle to the hair in order to minimize damage. After each sample was rinsed, the pantyhose was replaced, therefore minimizing the chance of cross-contamination. The hair was placed on a paper towel to dry and covered by a second paper towel. After the sample was air dried, it was placed into a new, labeled Ziploc bag.

Each case was then assigned a letter, "A" through "J," for identification. Five hairs per case were mounted in two different mediums on pre-cleaned glass microscope slides with glass cover slips. The proximal end of each hair, approximately one centimeter (cm), was cut and mounted on a microscope slide using Permount mounting medium (Fisher Scientific). The second cm of the same hair at the proximal end was cut and mounted on a microscope slide using Lactophenol Blue Stain (Dalynn Biologicals). Each hair was assigned a number in addition to its case letter. For example, the first hair examined in case "A" was called "A1," the second hair "A2," and so forth.

Three variables, having possible correlation with postmortem interval, were examined for each hair. The author examined each hair unaware of the PMI in order to reduce bias. Each hair was assigned a category for the amount of cuticle damage present, amount of fungal growth present, and the proximal end morphology (Table 2.1). For example, if a hair had moderate cuticle damage, no fungal growth, and root-banding present, then it would be assigned a 2 for cuticle damage, a 1 for fungal growth, and a 4 for proximal end morphology.

Table 2.1 Variable rating scales

Cuticle Damage:	
1	Little to no damage (normal wear and tear)
2	Moderate damage (slight lifting of the cuticle scales)
3	Severe damage (extensive lifting of the cuticle scales and loss of scales)
4	Absent cuticle

Fungal Growth:	
1	No growth
2	Little growth (fungal growth seen 1 or 2 times per cm)
3	Moderate growth (fungal growth seen 3 or 4 times per cm)
4	Extensive growth (fungal growth seen 5 or more times per cm)

Proximal End Morphology:	
1	Normal (usual antemortem anagen and catagen root)
2	Yellow-banding
3	Hard keratin point (Linch and Prahlow 2001)
4	Root-banding (Linch and Prahlow 2001)
5	Brush-like cortical fibers (Linch and Prahlow 2001)

The outer cuticle edge of the hair was observed in a longitudinal mount when assigning a hair to a category for cuticle damage. A hair was placed in category one for cuticle damage when the cuticle scales laid smooth and parallel with the shaft of the hair (Figure 2.1). A category of two for cuticle damage was assigned when the outer cuticle edge of the hair looked jagged and the distal tips of the scales appeared to be pulling away from the shaft of the hair (Figure 2.2). The cuticle may appear swollen, but was not essential for assigning the hair to category two for cuticle damage. A category of three for cuticle damage was assigned to a hair when the outer cuticle edge not only appeared jagged but some scales laid tangent to the shaft of the hair (Figure 2.3). The hair could have demonstrated a complete loss of scales on some areas of the shaft, but was not essential for assigning the hair a three for cuticle damage. Lastly, a category of four for cuticle damage was assigned to hairs in which the whole shaft was completely devoid of a cuticle layer. In this case the cortex was exposed and could appear to be fraying like a rope.

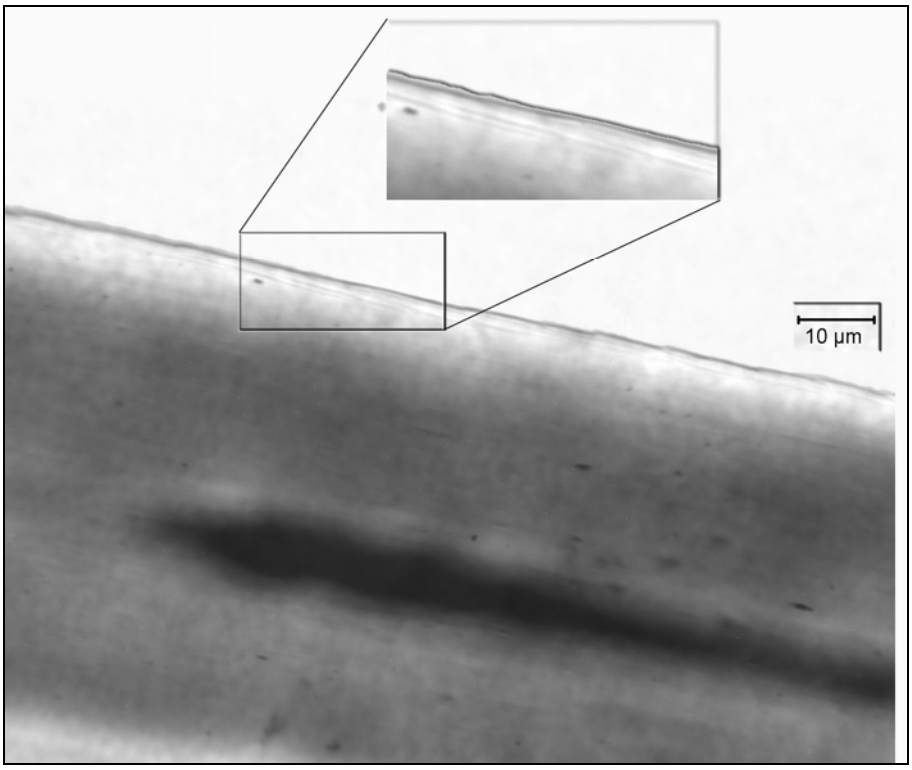

Figure 2.1 Transmitted light microscope digital image of control mounted with Permount mounting medium (example of cuticle damage 1: little to no cuticle damage)

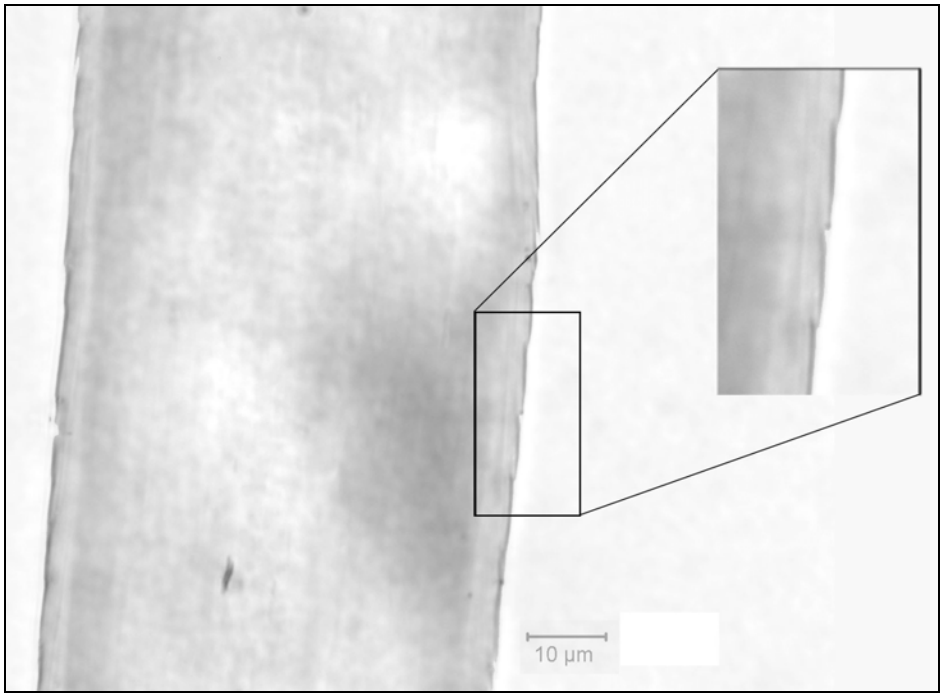

Figure 2.2 Transmitted light microscope digital image of putrid hair mounted with Permount mounting medium (example of cuticle damage 2: moderate cuticle damage)

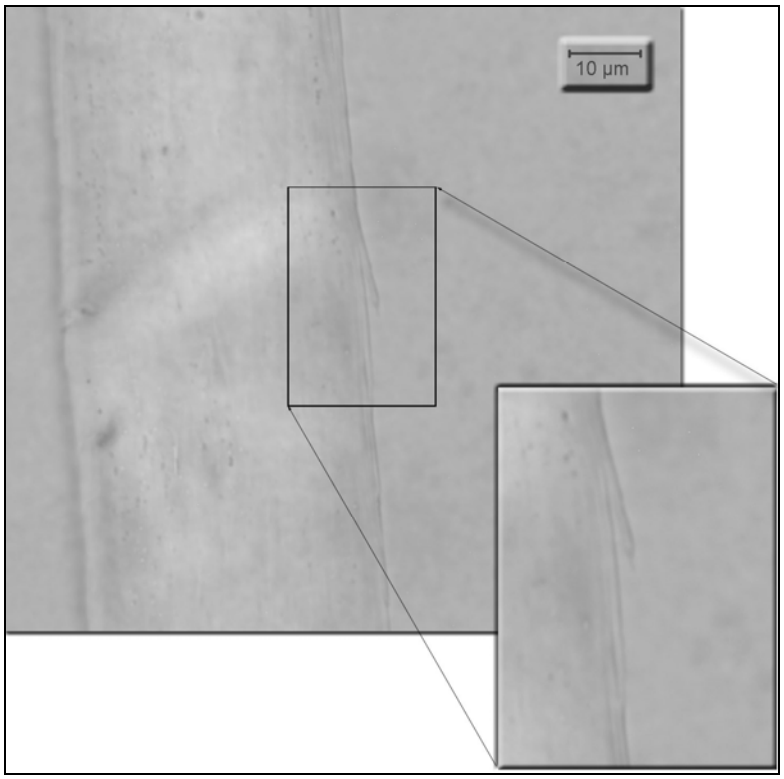

Figure 2.3 Transmitted light microscope digital image of putrid hair mounted with Permount mounting medium (example of cuticle damage 3: severe cuticle damage)

Due to the fact that not all fungi look alike or reproduce in the same manner, identifying specific fungal structures was not necessary to determine the category of fungal growth for each hair. Instead, I counted the fungal occurrences along one cm of the shaft of hair; no consideration was given to whether the fungal structures were vegetative or reproductive. A categorical value of one for fungal growth was assigned to hair that showed no evidence of fungal growth (Figure 2.4). The hairs that contained only one or two occurrences of fungal growth, no matter if the occurrence was a single hypha, mycelium, or fungal tunnel, were placed in category two for fungal growth (Figure 2.5). A hair was placed in category three for fungal growth if only three or four occurrences of fungal growth were witnessed along the shaft of the hair. Finally, a category of four for fungal growth was named if five or more occurrences of fungal growth appeared along the shaft of the hair (Figures 2.6 and 2.7). As stated earlier, the type of fungal structure witnessed did not play a part in the category of fungal growth assigned

for each individual hair. For example, even though the hairs in Figures 2.6 and 2.7 look different because of the type of fungus attacking each hair shaft, they are both placed in the same category of four for fungal growth.

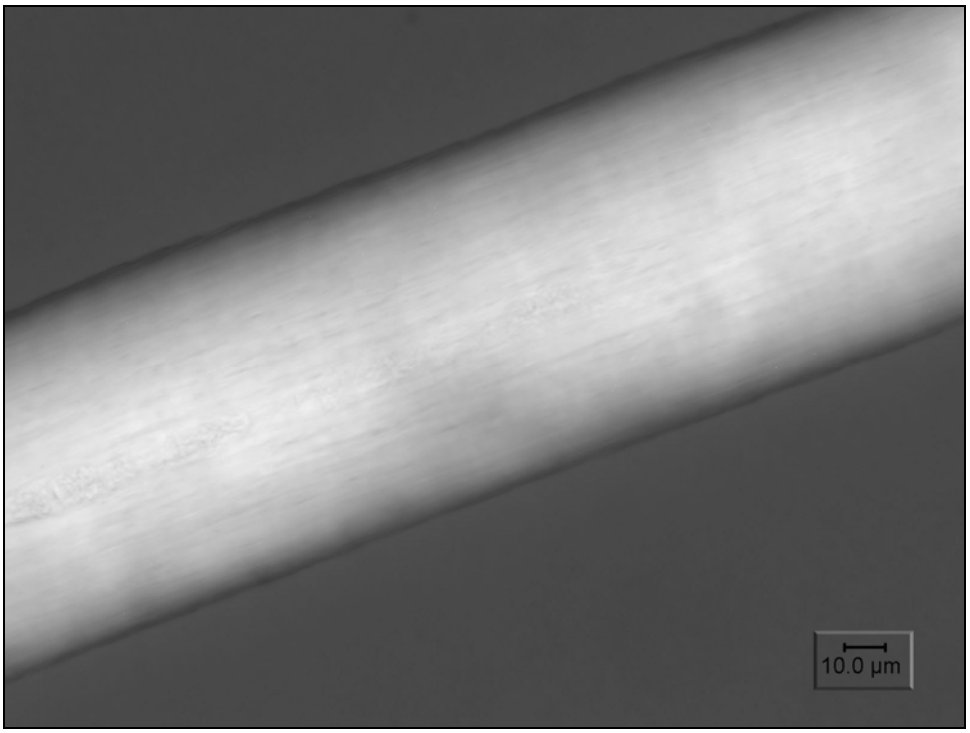

Figure 2.4 Transmitted light microscope digital image of control stained with Lactophenol Blue (example of fungal growth 1: no fungal growth)

The proximal tip and shaft of the hair were examined to determine the category for proximal end morphology. Hairs that looked similar to normal antemortem anagen and catagen roots of the hair were placed in category one for proximal end morphology (Figure 2.8). Hairs that had a yellow perpendicular band along the proximal end of the shaft were placed in category two for proximal end morphology. The descriptions and illustrations of hard keratin points, root-banding, and brush-like cortical fibers from Linch and Prahlow (2001) were utilized to assign hairs to categories three, four, and five for proximal end morphology. Hairs that had a root, which came to a "sharp" point were placed in category three for proximal end morphology (Figure 2.9).

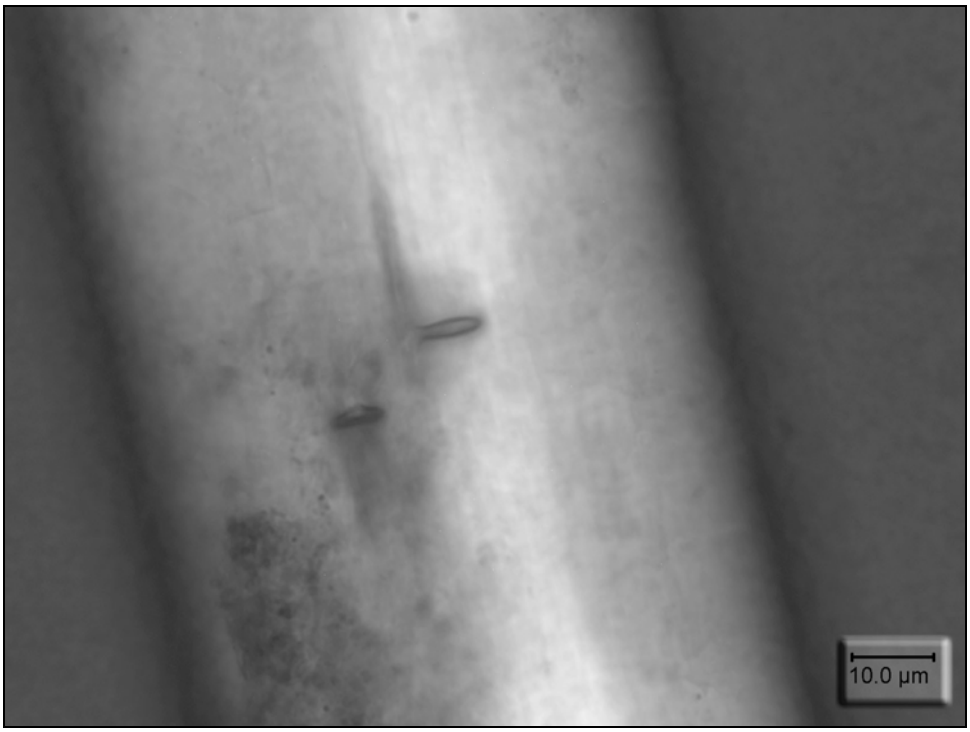

Figure 2.5 Transmitted light microscope digital image of putrid hair stained with Lactophenol Blue (example of fungal growth 2: little fungal growth)

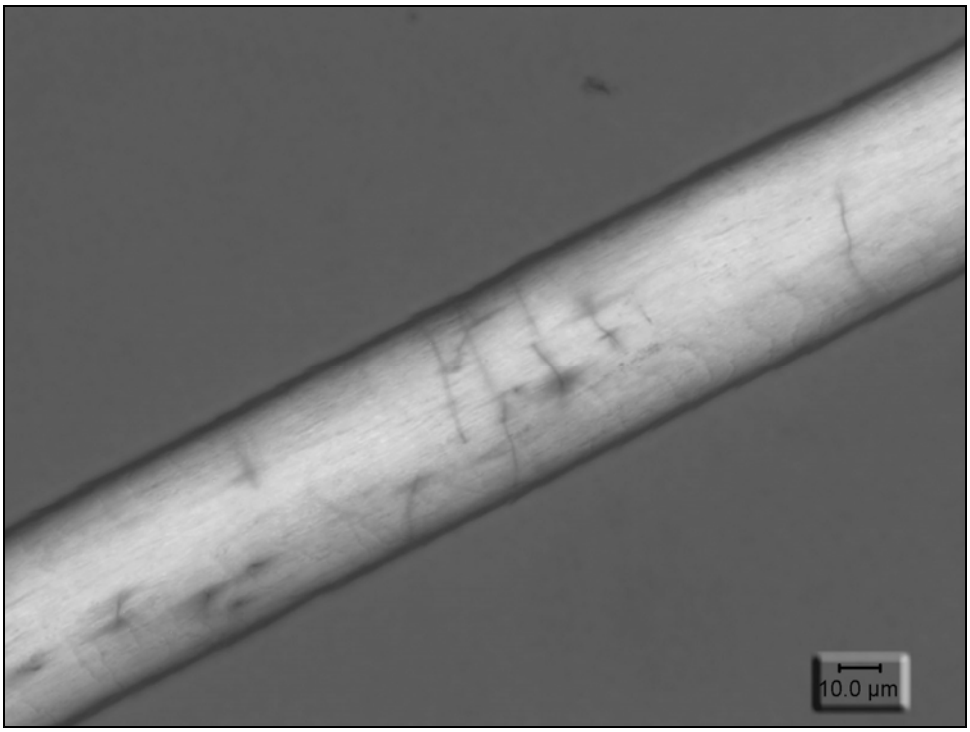

Figure 2.6 Transmitted light microscope digital image of fungal tunnels in putrid hair stained with Lactophenol Blue (example of fungal growth 4: extensive fungal growth)

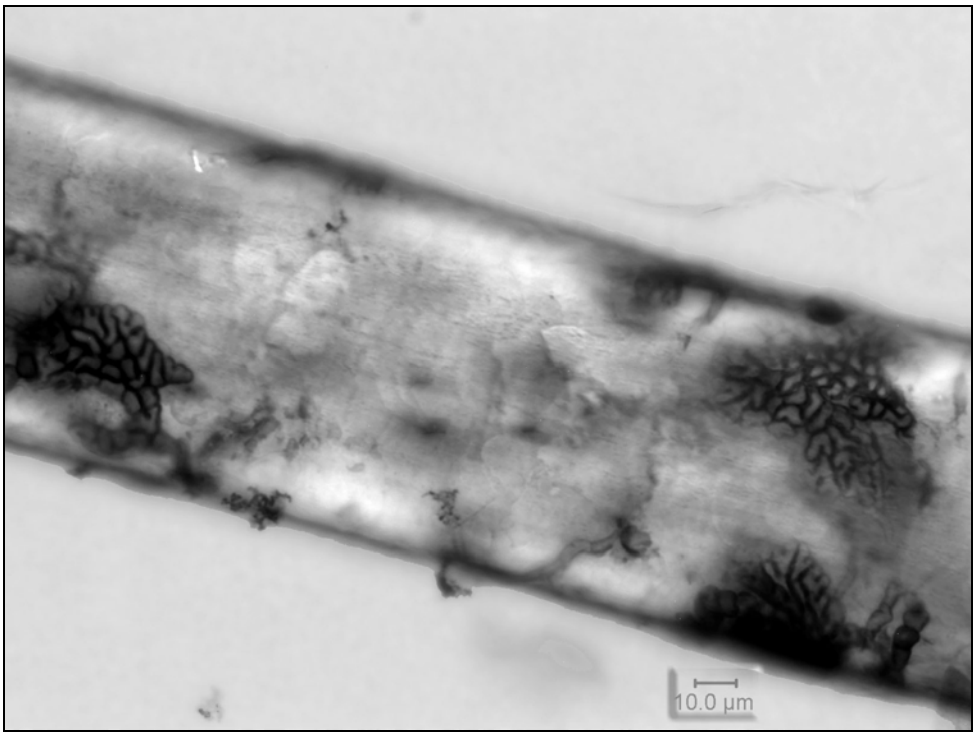

Figure 2.7 Transmitted light microscope digital image of putrid hair stained with Lactophenol Blue (example of fungal growth 4: extensive fungal growth)

Hairs that had a dark, perpendicular band along the proximal end of the shaft were placed in category four for proximal end morphology (Figure 2.10). Finally, hairs that had a brush-like proximal end were placed in category five for proximal end morphology (Figure 2.11).

The chi-square test was used in all statistical analyses, with the level of significance set at $P \leq 0.05$. SPSS software (2001) was used for statistical analysis and creation of graphs.

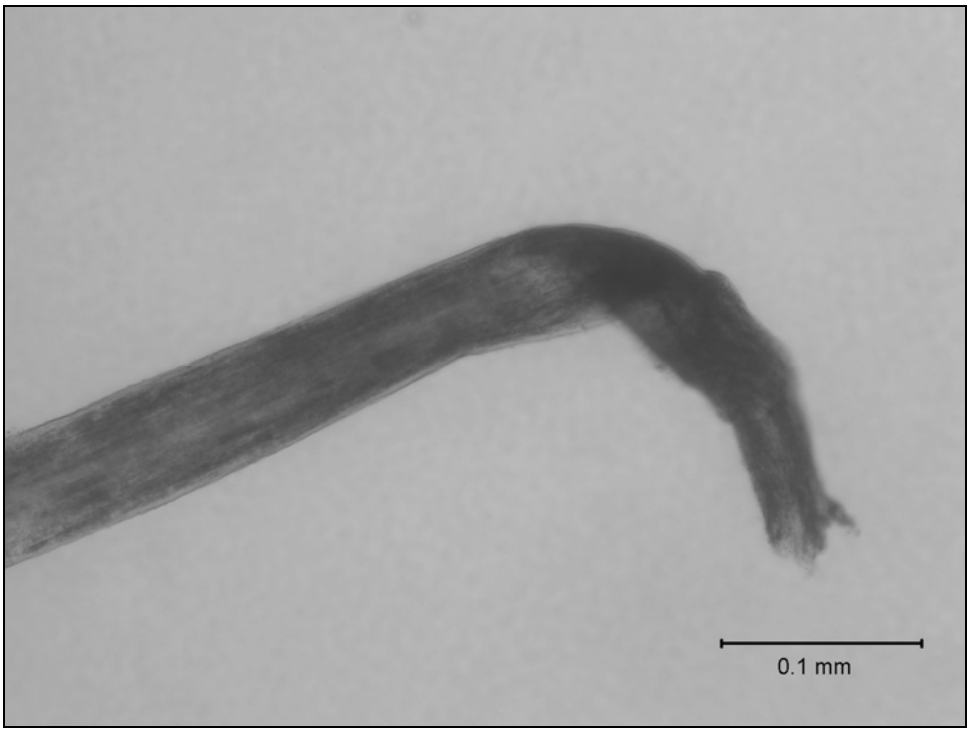

Figure 2.8 Transmitted light microscope digital image of putrid hair mounted with Permount mounting medium (example of proximal end morphology 1: normal antemortem root)

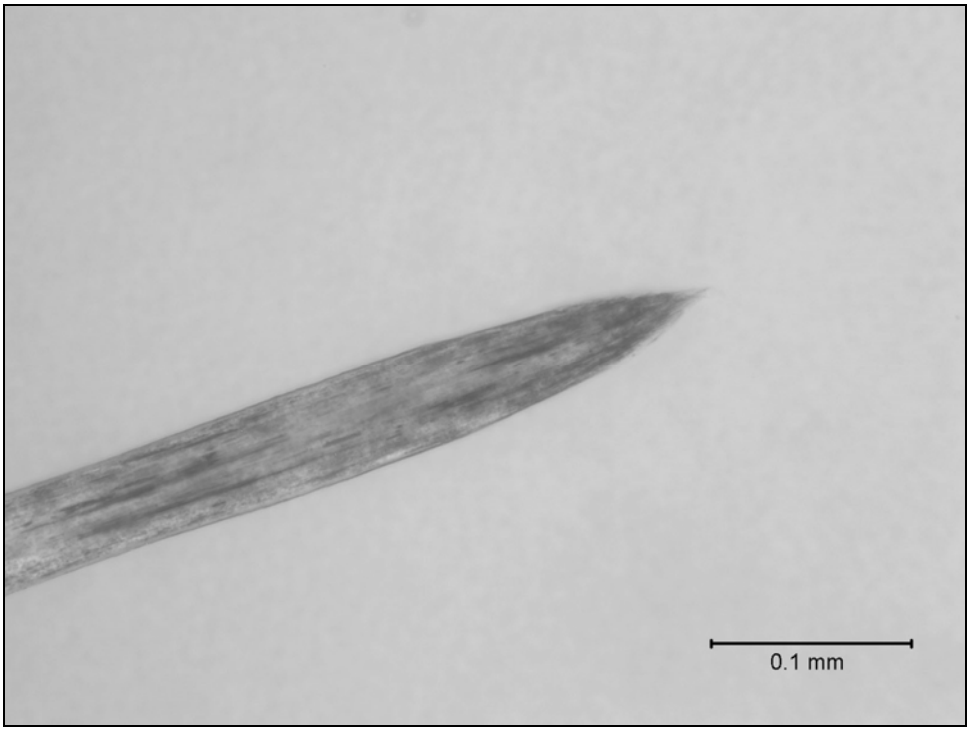

Figure 2.9 Transmitted light microscope digital image of putrid hair mounted with Permount mounting medium (example of proximal end morphology 3: hard keratin point)

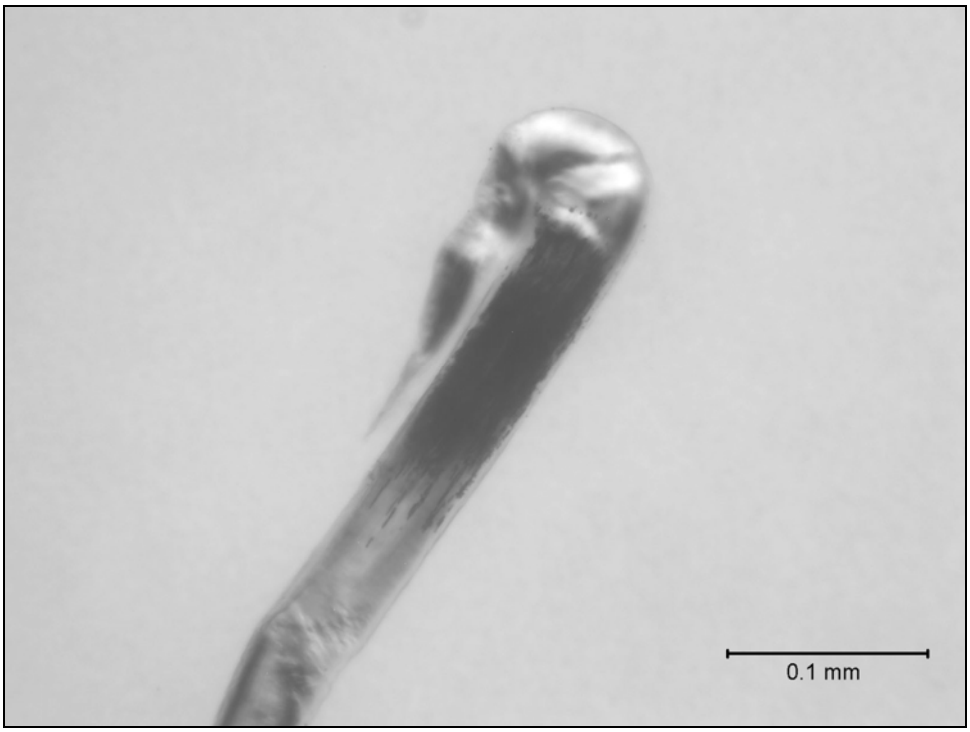

Figure 2.10 Transmitted light microscope digital image of putrid hair mounted with Permount mounting medium (example of proximal end morphology 4: root banding)

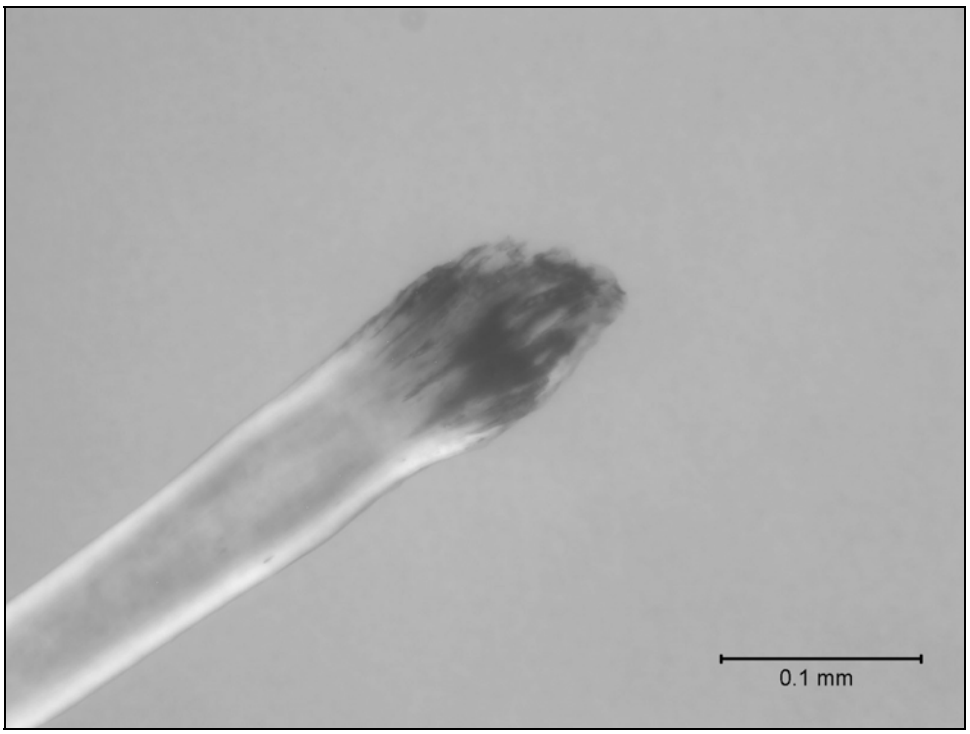

Figure 2.11 Transmitted light microscope digital image of putrid hair mounted with Permount mounting medium (example of proximal end morphology 5: brush-like cortical fibers)

CHAPTER 3: RESULTS AND DISCUSSION

Uniformity in Deterioration Rate

Before considering whether PMI is associated with cuticle damage, fungal growth, and proximal end morphology, it was essential to determine if all of the head hairs of an individual deteriorate at the same rate and not independently of one another. Twenty-five hairs from case "A" were examined for this analysis.

The null hypothesis that the cuticle of the hair from the same individual deteriorates independently of one another is rejected ($\chi^2 = 4.84$, df = 1, P = 0.028). Eighteen of the hairs, 72 percent, have category one cuticle damage. Seven of the hairs, 28 percent, have category two cuticle damage (Figure 3.1). No hair shows damage equivalent to categories three or four. In other words, the level of cuticle damage is predominantly that of category one.

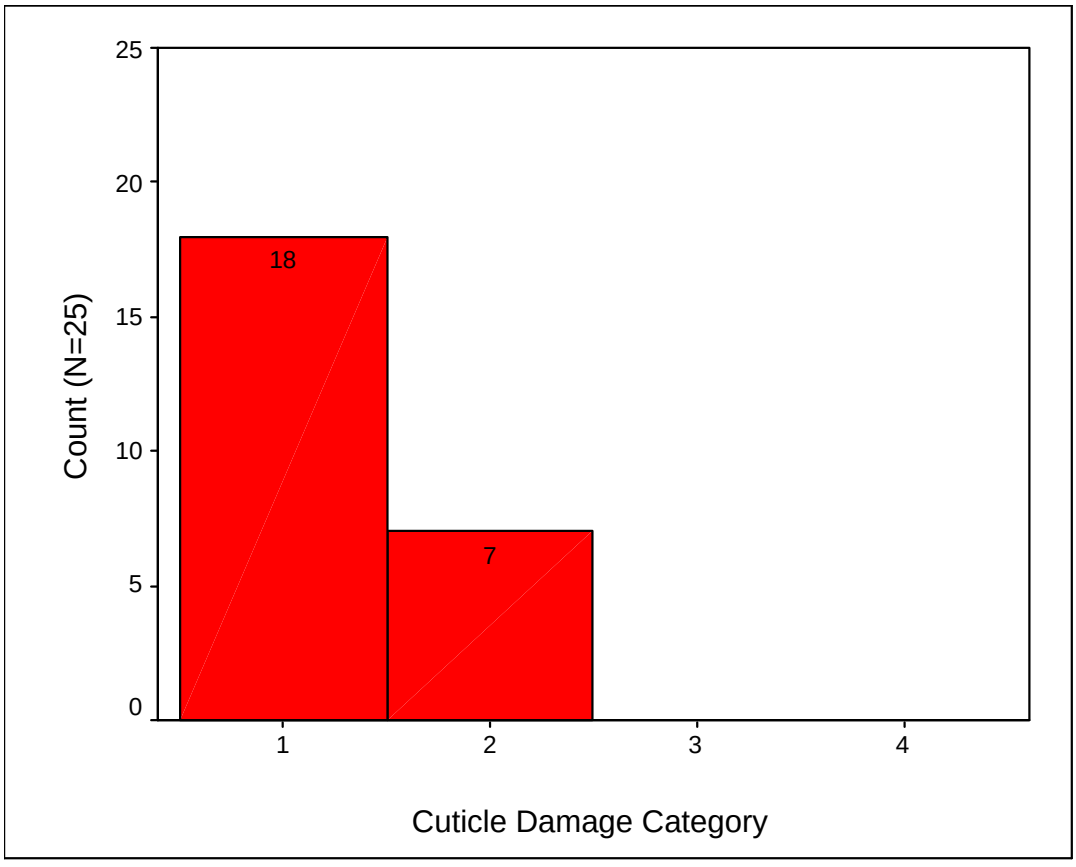

Figure 3.1 Case "A" count of cuticle damage categories

The null hypothesis that the proximal end morphology of the hair from the same individual changes independently of one another is rejected ($\chi^2 = 17.4$, df = 1, P = 0.001). Fourteen of the hairs, 56 percent, have category one proximal end morphology. Eight of the hairs, 32 percent, have category two proximal end morphology. One of the hairs, 4 percent, has category four proximal end morphology. Two of the hairs, 8 percent, have category five proximal end morphology (Figure 3.2). Therefore, hairs from the same individual show predominantly normal to yellow-banding in the proximal end.

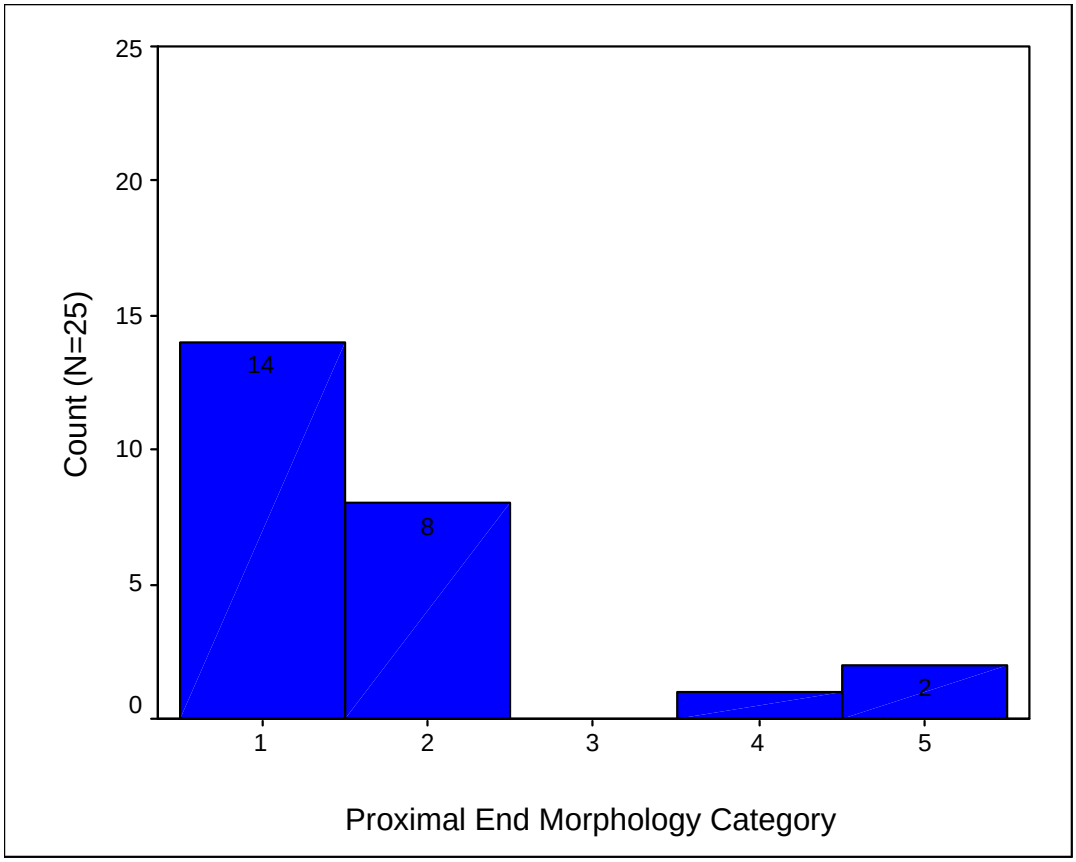

Figure 3.2 Case "A" count of proximal end morphology categories

Analysis of fungal growth shows that all 25 of the hairs, 100 percent, had category one fungal growth (Figure 3.3). Therefore, all hairs within an individual are consistent in showing no fungal growth.

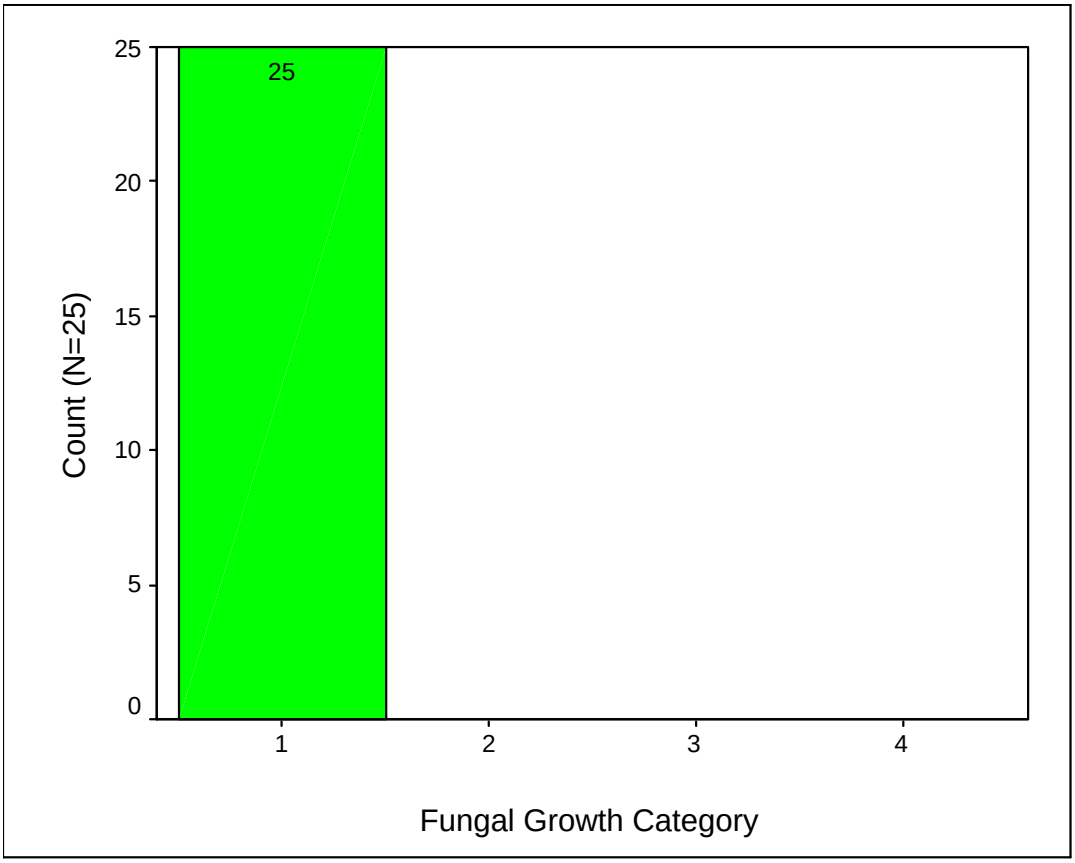

Figure 3.3 Case "A" count of fungal growth categories

Collectively, these results show that hairs from the same individual are generally similar to one another with respect to cuticle damage and proximal end morphology, and are identical to one another in fungal growth. In other words, each hair is generally representative of the sample as a whole. Therefore, one could postulate that the level of cuticle damage, type of proximal end morphology, and level of fungal growth are consistent on the head hairs of an individual at any given time. Because each of the five hairs in a case would be considered redundant, there was no longer a need to consider each hair individually, but to consider each case as a whole. Therefore, the sample size of 50 hairs cannot be considered to be independent, but rather to consist of ten individuals each with five (redundant) hairs. With this knowledge, a closer look could now be

taken at the deterioration rate of head hairs. This was achieved by comparing PMI with cuticle damage, fungal growth, and proximal end morphology.

Cuticle Damage, Fungal Growth, and Proximal End Morphology in Relation to PMI

Dissimilar to the statistical analysis of case "A" which had a sample size of 25 (i.e., 25 hairs), the statistical analysis of PMI with cuticle damage, fungal growth, and proximal end morphology was narrowed from four and five categories (Table 3.1) into two categories (Table 3.2) because of the small sample size (N=10; i.e., 10 individuals). Cuticle damage was narrowed into hairs having little to no cuticle damage and hairs having moderate to severe cuticle damage or absence of the cuticle. Each case was then assigned one of the two categories depending on the category where the majority of the five hairs were located. For example, case "C" had four hairs displaying some cuticle damage and one hair displaying no cuticle damage. Case "C" as a whole was then considered to have some cuticle damage. The same method of designation was used for separating fungal growth and proximal end morphology into two categories. Fungal growth was separated into hairs having no fungal growth and hairs having some fungal growth. Proximal end morphology was divided into hairs having normal and yellow-banding proximal end morphology versus hairs having a hard keratin point, root-banding, or brush-like proximal end morphology. Because each case possessed a different PMI, all of the cases were separated into one of two PMI categories. Cases with a PMI (PMI was considered to begin on date of deposition) of 90 days or less were considered to have a short PMI. Cases with a PMI of 91 days or more were considered to have a long PMI.

The relationship between cuticle damage and PMI is not significant ($\chi^2 = 1.27$, df = 1, P = 0.260). After taking a closer look at the cases involved, the author notices that the hair in case "H" displayed characteristics often associated with dyed hair, such as a clear demarcation line between dyed and undyed hair (Ogle and Fox 1999). According to Wolfram (2001), hydrogen

peroxide, which is used in the coloring of hair, leads to oxidative hair damage. Cases similar to "H", which involve dyed hair, could make the analysis of cuticle damage difficult because of the possibility that cuticle damage could have been present antemortem and there is no way of measuring how much damage was caused by hydrogen peroxide.

Fungal growth and PMI are significantly associated with one another ($\chi^2 = 10.0$, df = 1, P = 0.002). Unlike buried hair where fungi were evident after one month (Serowik and Rowe 1986; Kundrat and Rowe 1988), the first appearance of fungal growth in the present study occurred after 11 months. The difference in the time periods in which fungi first appear could be the result of different factors. The situation that Janaway (2002) described concerning the retarding ability of decomposing tissue on the deterioration of wool could play a role in the deterioration of hair. Also, all of the long-term studies of buried hair (Serowik and Rowe 1986; Kundrat and Rowe 1988) were conducted indoors in controlled environments, which could change the deterioration rate of hair. The field study from this experiment was outdoors where little control of the variables (precipitation, ultraviolet radiation, and temperature) could be exercised over the deterioration of head hair on the soil surface.

Table 3.1 Categorical assignments and PMI for cases A through J

		Category for		
Case	**PMI (days)**	**Cuticle Damage**	**Fungal Growth**	**Proximal End Morphology**
A	44	1	1	2
B	12	1	1	1
C	24	2	1	2
D	343	2	2	5
E	311	1	2	4
F	89	1	1	3
G	633	3	4	5
H	28	3	1	2
I	8	1	1	2
J (control)	0	1	1	1

Table 3.2 Condensed categorical assignments and PMI for cases A through J

		Category for		
Case	**PMI**	**Cuticle Damage Category**	**Fungal Growth Category**	**Proximal End Morphology Category**
A	short PMI	little to no cuticle damage	no fungal growth	normal and yellow-banding
B	short PMI	little to no cuticle damage	no fungal growth	normal and yellow-banding
C	short PMI	some cuticle damage	no fungal growth	normal and yellow-banding
D	long PMI	some cuticle damage	some fungal growth	hard keratin point, root-banding, and brush-like
E	long PMI	little to no cuticle damage	some fungal growth	hard keratin point, root-banding, and brush-like
F	short PMI	little to no cuticle damage	no fungal growth	hard keratin point, root-banding, and brush-like
G	long PMI	some cuticle damage	some fungal growth	hard keratin point, root-banding, and brush-like
H	short PMI	some cuticle damage	no fungal growth	normal and yellow-banding
I	short PMI	little to no cuticle damage	no fungal growth	normal and yellow-banding
J (control)	short PMI	little to no cuticle damage	no fungal growth	normal and yellow-banding

The relationship between proximal end morphology and PMI is significant ($\chi^2 = 6.43$, df = 1, P = 0.011). A type of proximal end morphology not previously described appeared in this study. Category two for proximal end morphology, the category for yellow-banding, was not originally one of the types of proximal end morphology researched in this study. The category was not added until observing several cases of proximal hair ends displaying this morphology. The numerous instances of yellow-banding seen at the proximal end of the hair could be the

result of the fact that many of the hairs examined in this study where white. In many cases, the yellow bands were difficult to see on white hair and, therefore, probably impossible to see on dark hair. Unfortunately, the yellow-banding seen in proximal end morphology could not be adequately seen in black and white photographs. Even though the reason for the yellow-banding at the proximal end is unknown, future research could study the possibility that the yellow-band is a precursor to root-banding. This possibility is based on the observation that yellow-banding was evident in cases with shorter postmortem intervals than cases with root-banding.

The postulation made by Petraco et al. (1988) that a brush-like cortical fiber could derive from the distal end of a broken root band agreed with the results of this study. Proximal ends that displayed brush-like cortical fibers were evident in cases with longer postmortem intervals. Also in agreement with Linch and Prahlow (2001), these cases could have arisen in the areas of the scalp which were moist.

Relationships among Cuticle Damage, Fungal Growth, and Proximal End Morphology

I evaluated the pairwise relationships among cuticle damage, fungal growth, and proximal end morphology. Similar to the tests using PMI, each case was placed in the category where the majority of its hairs fell. In addition, the four categories of cuticle damage and fungal growth and five categories of proximal end morphology were subsequently narrowed into two categories for each of the variables. The sample size (N) was ten, equivalent to the number of cases.

The relationship between cuticle damage and proximal end morphology was not significant ($\chi^2 = 0.278$, df = 1, P = 0.598). The relationship between cuticle damage and fungal growth was not significant ($\chi^2 = 1.270$, df = 1, P = 0.260). The association between fungal growth and proximal end morphology was significant ($\chi^2 = 6.429$, df = 1, P = 0.011). I suspect

that the relationship between fungal growth and proximal end morphology only exists because of the relationship they each share with PMI.

Final Remarks

Both fungal growth and proximal end morphology demonstrate a significant association with PMI. In contrast, cuticle damage is not significantly associated with PMI, perhaps because the cuticle is subject to antemortem change. Although proximal end morphology is associated with PMI, little is known about the chemistry behind the changes in proximal end morphology, and more research is needed. A limitation in applying fungal growth to estimating PMI is that fungal growth may vary among different climates.

The results of this study could be integrated into the current methods used in determining a PMI of a decedent found on the soil surface. The forensic examiner should collect 25 hairs from various areas on the decedent's scalp. The hairs should be lightly rinsed with water and allowed to dry. The proximal first cm of each hair should be mounted with a clear mounting medium such as Permount (Fischer Scientific). The second cm of the same hair at the proximal end should be cut and mounted with a stain, such as Lactophenol Blue, which is utilized in mycological examinations. A compound light microscope is necessary to examine both types of slides. The presence of a normal root or yellow-banding on the proximal end and no fungal growth on a head hair would suggest a PMI of < 90 days. In contrast, the presence of a hard-keratin point, root-banding, or brush-like proximal end and fungal growth on a head hair would suggest a PMI of > 90 days. Utilized in conjunction with other dating methods, the observations of fungal growth and changes in proximal end morphology of human head hair may prove beneficial in estimating a PMI.

Future research in human head hair deterioration rates would be to determine the broader applicability beyond the site-specific results of this study. Furthermore, examining the

relationships of fungal growth and proximal end morphology with PMI in a study with a larger number of cases over three months old would give a better understanding of the intermediate PMI. A logical follow-up project to this research would be a long-term study, which examined the deterioration rate of head hair (still connected to the scalp) in different environmental conditions, such as buried, submerged, and on the ground surface. Future research in these areas would make it possible to construct a method to estimate postmortem intervals in cases of unknown time since death.

CHAPTER 4: CONCLUSION

This study found that head hair from the same individual deteriorates uniformly. Furthermore, fungal growth and changes in proximal end morphology were found to have a significant association with PMI. Cuticle damage, on the other hand, was found to have a nonsignificant relationship with PMI. The relationships between cuticle damage and fungal growth, and cuticle damage and proximal end morphology were not significant. In contrast, there was a significant association between fungal growth and proximal end morphology.

Further research is needed to give a more complete picture of the relationship between human head hair deterioration and PMI. Studies consisting of longer postmortem intervals, with a larger number of cases, would be useful. In addition, experiments that expose hair (still associated with the scalp) from the same decedent to different environments and climates could aid in the understanding of the universal deterioration rates of human head hair.

The author suggests that during the forensic investigator's examination of a decedent with an unknown PMI, a sample of 25 head hairs should be collected and saved for evaluation. The slow decomposition rate of hair, relative to other soft tissues, makes it a valuable source of information in older forensic cases. Utilized in conjunction with other dating methods, the observations of fungal growth and changes in proximal end morphology of human head hair may prove beneficial in estimating a PMI.

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APPENDIX

CASE BACKGROUND INFORMATION

Case	Age	Sex	Ethnicity	Cause of Death	Date of Death	Date of Deposition
A	59	F	Caucasian	diabetic/breast cancer	6/20/2003	6/21/2003
B	62	M	Caucasian	lung cancer	7/20/2003	7/23/2003
C	77	M	Caucasian	lung cancer	7/8/2003	7/10/2003
D	63	M	Caucasian	cardiopulmonary disease	8/26/2002	8/26/2002
E	64	M	Caucasian	unknown	9/26/2002	9/27/2002
F	55	M	Caucasian	unknown	4/20/2003	5/9/2003
G	76	M	Caucasian	myocardial infarction	11/9/2001	11/9/2001
H	52	F	Caucasian	unknown	7/6/2003	7/7/2003
I	71	M	Caucasian	myocardial infarction	7/26/2003	7/29/2003
J (Control)	59	M	Caucasian	N/A	N/A	N/A

VITA

Jamie Hughes Collier was born and raised in Harvey, Louisiana. She received her Bachelor of Science degree in biology with a minor in criminal justice from Northwestern State University (NSU) in Natchitoches, Louisiana, in May 2001. While at NSU, Jamie was named the Northwestern State University Student Leader of 2000. She was married in the summer of 2001 and subsequently moved to Baton Rouge, Louisiana, with her husband. She began graduate school in the fall of 2001 at Louisiana State University (LSU) and will be graduated in the spring of 2005 with a Master of Arts degree in anthropology. While at LSU, Jamie was a Co-President of the Geography and Anthropology Society and a student affiliate of the American Academy of Forensic Sciences. She plans to continue her research in human hair biodeterioration and pursue a career in the forensic sciences.

rejected counts were caused by high DAPI or FITC background in that area and red dots (positive in FITC but negative in DAPI). Most false positives in the image analysis software were quickly rejected by visualizing the gallery (captured cells) on the computer screen. Ambiguous signals/cells were accepted or rejected by visual examination using both the FITC and DAPI filters. Manual scoring of human spermatozoa and the setting up of the image analysis software took the same amount of time. While the image analysis software carries out the automated scoring of human spermatozoa, other tasks can be performed. A major advantage when counting multiple slides is the elimination of eye strain as reviewing galleries shown on the large computer screen is an easy and quick step.

The results of this study indicate that automated scoring of fluorescently-stained human spermatozoa in mock sexual assault exhibits can be carried out reliably and reproducibly using well-developed classifiers for the image analysis software system. The automated scoring of spermatozoa combining the fluorescence-based staining assay and the image analysis software is currently being tested on a large number of sexual assault cases as part of a pilot project within an operational setting.

Automated, Spermatozoa, Image Analysis Software

A157 Re-Evaluation of the Seratec® PSA Semiquant Test for Use at the UnitedStates Army Criminal Investigation Laboratory

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After attending this presentation, attendees will be educated regarding the detection of PSA using the Seratec® PSA Semiquant test on samples and how the United States Army Criminal Investigation Laboratory (USACIL) has addressed low levels of PSA present in casework.

This presentation will impact the forensic science community by increasing awareness about false positive PSA results using the Seratec® PSA Semiquant test when performing the test on evidentiary samples. Knowing what substances can be attributed to false positive reactions to the Seratec® PSA Semiquant test, and how to dilute them from the sample without compromising the examiner's ability to identify semen, would help to improve confidence in the results that are presented in court.

It is known that semen contains a high concentration of PSA, making PSA a useful biological marker to identify semen. The Seratec® PSA Semiquant test has been determined to be a valid and reliable method for detecting semen in biological stains. The test works by detecting PSA using two monoclonal antibodies that combine with the PSA to form a complex which is visualized as a red line on a membrane. The sample is extracted in a buffered solution to maintain a constant pH and to help it travel through the test strip. It is also well documented that a small chance of false positive PSA results in the absence of semen. Sometimes these results are from elevated PSA due to a biological phenomenon in an individual. Other times this is a result of non-biological material mimicking a positive result on the test strip. In the literature it has been noted that a change in pH due to the addition of organic acids (citric acid, acetic acid, and oxalic acid) can cause a false positive band on the Seratec® PSA Semiquant card. Examiners at USACIL perform Acid Phosphatase (AP), PSA, and microscopic examinations on a sample to determine if semen is present. Sometimes all three tests are performed on the same cutting. If a sample is only PSA positive an immunological indication of semen is reported. A review of cases at the USACIL has noted the presence of weak positive PSA results with no male DNA detected in multiple samples from different cases. In order to determine if a case sample is truly positive for semen, a study was performed to test various non-biological samples in order to isolate any that may show a false positive PSA result. Diet soda, alcoholic beverages, tooth paste, lubricants, douche, mouthwash, and various other substances were tested. During preliminary studies it was found that diet sodas gave false positive results when diluted one to one in PBS. The effects of AP reagent on the Seratec® PSA Semiquant test were also examined to determine if there was any interference occurring. Various dilutions were performed to determine if any changes to the current USACIL protocol can help eliminate these false positive results while still being able to detect low levels of PSA from semen.

Seratec®, PSA, False Positive

A158 The Microscopic Characteristics of Antemortem and Postmortem Hairs at the Root End

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After attending this presentation, attendees will see how the microscopic effects various environmental conditions may have on hairs that have been removed antemortem. This presentation will also demonstrate how to microscopically distinguish these hairs from hairs that have undergone postmortem changes.

This study will benefit the forensic science community by providing information on the microscopic characteristics that may be caused by environmental exposure and how these characteristics differ from postmortem banding. Postmortem banding and putrid roots are microscopic characteristics commonly observed in hairs that have undergone postmortem changes. Based on the experience of hair examiners, postmortem banding is generally accepted throughout the forensic hair community as a reliable indication of hair removal during the postmortem process. However, few research studies have been conducted to address the possibility that these characteristics may be observed in hairs removed antemortem.

Results from a study involving 600 hairs collected from fifteen living individuals will be presented. These hairs were exposed to various environments including indoors on a windowsill, submerged in water, buried in potting soil, outdoors on the ground surface, and inside vehicles. The hairs were subsequently microscopically examined at the root end to determine the type of changes that may have occurred as a result of storage in these conditions. Any changes at the root end were then compared to hairs removed from deceased individuals.

The majority of hairs studied (97%) contained roots in the actively growing anagen phase. Hairs in the anagen phase are not completely keratinized and thus more susceptible to changes due to environmental conditions. Two-hundred and fifty hairs were stored in vehicles or indoors on a windowsill for time periods ranging between nine days and 230 days. Two-hundred and fifty hairs were stored outdoors on the ground surface in shaded and non-shaded areas for time periods ranging between seven days and 106 days. One-hundred hairs were submerged in water or buried in potting soil for time periods ranging

between fifteen days and 100 days. No hairs stored indoors on the windowsill and no hairs stored in the vehicles exhibited characteristics of decomposition. Some of the hairs stored outdoors, most of the hairs submerged in water, and most of the hairs buried in potting soil exhibited characteristics of decomposition. Some of these characteristics are similar to characteristics observed in hairs removed postmortem. However, no hairs in this study exhibited characteristics of postmortem banding.

The results from a blind test on the identification of postmortem banding will be presented. The test consisted of over 200 hairs that included all hairs in the presented study with possible changes at the root end as well as hairs known to have been removed from deceased individuals. Initial analysis by the two examiners that completed the test resulted in greater than 99.5% accuracy in identifying postmortem banding. Following a confirmation process whereby each examiner reviewed the initial results, accuracy was increased to 100%.

Results from this study contribute towards the reliability that hairs exhibiting characteristics of postmortem banding are consistent with having been removed postmortem. This study also demonstrates the need for proper training and good quality assurance procedures when identifying postmortem banding.

Hair, Postmortem, Banding

A159 The Evaluation of Possible False Positives With Detergents When Performing Amylase Serological Testing on Clothing

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After attending this presentation, attendees will know that false positive detection of -amylases is not possible when testing clothing laundered in detergents containing a-amylases to screen for the presumptive presence of saliva on evidentiary clothing.

This presentation will impact the forensic science community by increasing the understanding of presumptive saliva screening methods, and will clarify any misconceptions between the scientific and legal communities regarding the sensitivity and specificity limitations of the Phadebas® and RSID™-Saliva screening methods employed for the detection of α-amylase on evidentiary samples.

Amylase detection has become a very useful tool in the screening process for possible saliva stains on forensic evidence. In particular, α-amylase detection using commercially available tests such as RSID-Saliva or Phadebas® Amylase Test are two ways in which the forensic scientist is able to determine if saliva is presumptively present on an evidentiary sample.

The enzyme α-amylase is naturally occurring in many species of bacteria, fungi, plants, and animals. As a result, the presence of two α-amylase isoenzymes in humans has led to some difficulty in reporting the presence of salivary α-amylase versus pancreatic α-amylase in forensic casework. Moreover, there has been recent speculation from legal professionals that the α-amylases present in common household laundry detergents may be contributing to positive detection of α-amylase on evidentiary samples during forensic presumptive screening procedures.

For almost 40 years, detergent companies have been adding enzymes such as proteases, celluloses, and amylases to their products as a more effective method of breaking down tough stains created by polysaccharides and proteins. To determine whether or not α-amylase detection is possible following routine clothing laundering, unworn, unwashed fabrics of different compositions were laundered in a variety of detergents and stain removing agents. Two assays, RSID™-Saliva (Independent Forensics, Hillside, IL) and Phadebas® Amylase Test (Magle Life Sciences, Lund, Sweden), that use different methods of detecting α-amylase, were used to investigate whether detergent α-amylases in laundered clothing are detectable. RSID-Saliva detects human salivary α-amylase via anti-salivary amylase monoclonal antibodies whereas the Phadebas® assay takes advantage of the enzymatic properties of α-amylase present in a given sample. Thus, when using a screening method such as the Phadebas® Amylase Test, a positive amylase result is only suggestive, and not confirmatory, for the presence of human saliva.

Five fabric swatches were washed in a volume of laundry detergent pre-determined for a light load wash cycle, exceeding what would typically be required to launder five fabric swatches. This was employed in an attempt to maximize the retention of detergent and detergent additives in the laundered clothing. Nevertheless, all garments tested negative in response to alternate light source, Phadebas®, and RSID-Saliva. These findings suggest that at some point during the laundering cycle, the enzymes were damaged, degraded, or removed. The causes of a-amylase loss during laundering were not examined; however, the data support that detergent enzymes should not contribute to a misidentification of a saliva stain using a presumptive screening method.

Additionally, unlike laundered clothing, undiluted detergents do contain detectable levels of α-amylase, but these findings were only observed using the Phadebas® Amylase Test.

Amylase, Saliva Screening, Serology

A160 Capturing the Moment: Photographing Low Level Signals From Serological Testing of Swabs and Cartridges

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After attending this presentation, attendees will have learned how to photograph, in a single image, multiple DNA swabs, or cartridge tests with varying levels of signal intensity from presumptive color tests or from cartridge tests for blood or semen.

This presentation will impact the forensic science community by providing an optimal way to photograph the results of time-sensitive Serological tests of varying signal intensities in order to document the findings of a Forensic Genetics analyst.

To photograph the results of presumptive color tests on swabs or the results of blood or semen cartridge tests, the photographs must be taken at specific time interval after the test is completed. The photograph must accurately capture what the analyst observes even when the results are faint. The Forensic Imaging Division and the Forensic Genetics Laboratory of the Harris County Institute of Forensic Sciences conducted a study to establish photographic standards and procedures that would accurately capture and corroborate the results observed by the analyst within the time constraints. This interdisciplinary effort was designed to identify and validate a photographic method so that the images could be used for verification of the analyst's work. These results are intended to provide guidance to other agencies in establishing and validating photographic methods in order that photographs may become a standard and useful part of the case record.

A Nikon D5000 camera with a Nikkor 85mm macro lens was used to perform the tests with an ISO of 200, a shutter speed of 1/125th of a second and an aperture setting of *f*/22. Due to the fact that several swabs or cartridges are often collected at the same time, these settings had to be able to accurately portray all levels of signal intensity in a single shot. For the swabs, a series of tests on various colored backgrounds, exposure settings, and lighting angles was conducted. Settings were validated by changing one variable at a time and holding the others variables constant. Successful results were observed at -0.3 exposure compensation on an 18% neutral gray background with direct lighting. This proved to yield an image that captured a wide range of signal levels on several different swabs within a single image.